CERTIFICATE

This is to certify that the dissertation titled **"Hepatotoxicity in patients receiving HAART therapy: Clinical Profile & Risk factors"** is a genuine work done by **Dr. P. Karthikeyan**, Post graduate in Medical Gastroenterology under my supervision between June 2006 to February 2007 and is being submitted in partial fulfillment of the requirement for the awarding of **D.M. (Medical Gastroenterology)** degree by the Tamil Nadu Dr. M.G.R. Medical University, Chennai.

Dean
Madras Medical College,
Chennai – 600 003

Prof. S. Barnaba Durairaj, MD., DM
Department of Medical Gastroenterology
Madras Medical College,
Chennai – 600 003

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CONTENTS

1. INTRODUCTION

HIV infection/AIDS is a global pandemic with cases reported from virtually every country. The current estimate of cases among adults is estimated to be around 37 million. Two thirds of them live in Sub Saharan Africa and 50% are women. The prevalence ranges from 0.6% to 8% in different parts of the world.[1]

Gastroenterological manifestations are quite common in advanced HIV infection/ AIDS and are some times initial manifestations or AIDS defining illness. Liver involvement is unique since it can be because of direct involvement, opportunistic infections (OI) with r co-infection of one or more hepatotrophic viruses, or because of toxicity due to retroviral drugs.[2]

HEPATIC MANIFESTATION OF HIV/AIDS

Presence of liver disease is a frequent finding in AIDS. Hepatomegaly may be detected on examination in most patients. Hepatomegaly is usually associated with one or more liver chemistry test abnormalities, although significant jaundice due to parenchymal disease is uncommon. As with other organ systems, the spectrum of hepatic infections in patients with HIV evolves as immunocompromise advances. Clinical manifestations of hepatobiliary disease can vary from no symptoms to liver failure.

Hepatobiliary disease can be broadly classified into either hepatic parenchymal abnormalities, biliary abnormalities, or a combination of the two. Currently, parenchymal abnormalities are most often related to viral hepatitis and drug-induced disease; however, neoplasms, in particular non-Hodgkin's lymphoma, and OIs are not infrequent

Drug-induced liver injury has emerged as the most prevalent cause of liver test abnormalities and is related to the increasing array of antiretroviral medications. Use of prescription or nonprescription drugs as well as herbal remedies should also be considered a cause of abnormal liver test results in the HIV-infected patient. Before HAART, drug hepatotoxicity was most commonly due to sulfonamides, and the increased frequency of adverse reactions to these medication is well recognized in AIDS.

Well established syndromes characterized by marked hepatomegaly, steatosis, lactic acidosis, and liver failure has been increasingly recognized.Hepatic steatosis is usually evident on imaging of the liver. Although reversal has occurred in some patients following drug withdrawal, most patients have worsening disease and death. Liver transplantation is curative.

The incidence of drug induced hepatotoxicity, its mechanisms, evaluation, management, analyses of various studies on the risk factors and prognosis are described in detail in literature review. Other causes of liver dysfunctions are reviewed below and should be kept in mind before evaluating these patients.[2]

INFECTIONS

Mycobacterium avium intercellulare complex (MAC) is consistently the most frequent specific hepatic finding in AIDS, documented in up to 46% of patients in late-stage HIV disease. The pathologic hallmark of the infection is the presence of poorly formed granulomas containing acid-fast bacilli within foamy histiocytes. Organisms may be observed in the absence of granulomas and can be cultured from liver biopsy in the absence of infected histiocytes

Mycobacterium. tuberculosis, in contrast to MAC, may occur before HIV-infected patients are profoundly immunocompromised. Extrapulmonary tuberculosis is common in patients with HIV infection, especially in patients with prior OIs and those whose risk behavior is injection drug use. Hepatic disease as part of miliary tuberculosis has been noted. Rarer manifestations include tuberculous abscesses and bile duct tuberculomas. The diagnosis of hepatic tuberculosis is made by culture of the organism from liver tissue obtained by percutaneous or laparoscopic biopsy. PCR may be helpful.

CMV is the most frequent infectious pathogen in AIDS, and the liver is involved in 5% to 25% of liver biopsies. However, its discovery in the liver antemortem is extremely unusual. Typical viral inclusions are usually identified in Kupffer cells .

Clinical manifestations and histologic features of *viral hepatitis* from HBV, HCV, or hepatitis D virus are altered in the presence of HIV coinfection but in remarkably different ways for each virus.

Clinical and autopsy studies in AIDS patients have reported up to a 90% seroprevalence of hepatitis B markers indicating past or present infection.

Concurrent HIV and HBV infections lead to alterations of HBV antigen-antibody display, viral replication, and clinical consequences. Several reports have described reappearance of hepatitis B surface antigen (HBsAg) in HIV-infected patients previously thought to be immune to hepatitis B virus as indicated by the presence of anti-HBs.[5]

Recurrence of HBsAg may arise from either reinfection or reactivation with advanced immunodeficiency. In addition, there is an accelerated loss of naturally acquired anti-

HBs even in those patients who remain HBsAg negative. With loss or reduction in immunity to HBV, there is an increased prevalence of hepatitis B e antigen expression, elevated mean levels of DNA polymerase, and increased titers of anti–hepatitis B core antigen.[6]

Acquisition of the chronic carrier state is also much more likely in the HIV-infected patient, especially if infection occurs when immunodeficiency is more advanced. Thus, a larger proportion of patients with HIV and hepatitis B infections have a chronic carrier state, with highly infectious serum and body fluids, compared with those who are HIV negative

Although HIV infection leads to more prevalent chronic HBV carriage, it appears to attenuate the severity of biochemical and histologic liver disease in most, but not all[1] patients. In one study, the mean alanine aminotransferase (ALT) level correlated with CD4 lymphocyte count. The mechanism for reduced hepatitis B virus–related liver injury following HIV infection is not certain but has been attributed to a diminution in lymphocyte-mediated hepatocellular injury as a result of HIV effects on lymphocytes. In those patients without serologic evidence of past or present hepatitis B virus and HIV infection, vaccination appears to be ineffective, regardless of the stage of immunocompromise.

Sometimes the institution of HAART in a chronic carrier of hepatitis B virus can have catastrophic consequences. Patients may develop an acute flare of hepatitis that can be severe leading to fulminant hepatic failure. However, the proportion of coinfected patients who develop an acute hepatitis B flare following use of HAART is unknown. It is believed that reconstitution of immune function with HAART leads to production of antibody that is directed to infected hepatocytes as in the normal host. Inclusion of

lamivudine, which has potent antiviral effects on hepatitis B virus, in the HAART regimen may reduce the likelihood of acute hepatitis B.[6]

The consequences of HIV infection on delta hepatitis appear similar to those of HBV, although far fewer patients have been studied.

The prevalence of HCV in those with HIV infection depends on the risk group evaluated and the assay used. Prevalence is highest in injection drug users (52% to 89%)[7,8] and hemophiliac patients with HIV, whereas in military populations and non–drug users, the prevalence is much lower, ranging from 1% to 11%. Assaying antibodies to hepatitis C virus alone, rather than hepatitis C virus RNA, may underestimate the true prevalence, because loss of antibody may occur with progression of immunodeficiency.5, 6

Unlike hepatitis B virus, the clinical course of hepatitis C virus appears to worsen as HIV-related immunocompromise advances. This has been best documented in HIV-infected hemophiliac patients. Studies in large cohorts of hemophiliac patients have demonstrated dramatic increases in hepatitis C virus RNA levels with progressive HIV disease, associated with aspartate aminotransferase (AST) elevations and hepatomegaly. Co infected patients also have a higher rate of active cirrhosis on biopsy and an accelerated course to clinical cirrhosis and liver failure. The mechanism for this more rapid disease course is unknown but has been similarly recognized in other immunocompromised patients. Because patients with late-stage HIV often have multiple life-threatening infections, HCV alone is not an independent determinant of mortality. However, as HIV-infected patients are living longer owing to HAART, hepatitis C virus–induced liver disease and its consequences (e.g., hepatocellular cancer) are assuming more clinical relevance. Like hepatitis B, hepatitis C virus does not cause progression of HIV disease. [7]

The effect of HAART on hepatitis C viral dynamics and liver injury is variable. Some studies have found attenuation of disease, whereas others had documented exacerbations reflected by increases in serum transaminases. Hepatitis C viral load has also been variably affected. [8]

The role of interferon therapy for HIV/HCV coinfected patients remains unsettled. α-Interferon is less effective for treating hepatitis C virus liver disease in coinfected patients . More recently, combination therapy of Peg-interferon and ribavirin has shown promise. [10]

Fungal infections of the liver are not unusual when immunocompromise is advanced. *Histoplasmosis* of the liver may be seen in patients with disseminated fungal disease, predominantly but not exclusively in regions of high prevalence of the organism. Biopsies of the liver may also show caseating granulomas containing fungal organisms. Culture of hepatic tissue, blood, or bone marrow can confirm the diagnosis, but several weeks may be required for the organism to grow in culture.

Pulmonary disease is seen in most patients at diagnosis.*Cryptococcus* may infect the liver in the setting of disseminated infection. Typically the organism is found in the sinusoids and is associated with a poor inflammatory response. Similarly, coccidioidomycosis can involve the liver as part of a systemic infection, especially in endemic regions. The organisms appear as spherules within a fibrosing granulomata.*Candida* infection of the liver is rare, in contrast to its high prevalence in mucosal sites. Hepatic microabscesses or macroabscesses are most likely to occur if the patient is neutropenic, especially following chemotherapy for non-Hodgkin's lymphoma.

Kaposi's sarcoma, which is caused by infection with human herpesvirus 8 (HHV-8) has a predilection for periportal regions of the liver and is seen in 10% to 15% of liver biopsies.

Tumor nodules appear grossly as violaceous or hemorrhagic masses within hepatic parenchyma. Microscopically, the characteristic spindle cells and vascular slits of Kaposi's sarcoma usually directly abut normal-appearing liver tissue.

Hepatic involvement by *non-Hodgkin's lymphoma* may be the index manifestation of AIDS in homosexual men and may be the primary site of the neoplasm. The lesions are usually focal and may be large. In addition, Hodgkin's disease in the AIDS patient tends to be more aggressive histologically and clinically, spreading rapidly to extranodal sites, making liver involvement more likely.

Isolated cases of *P. carinii* pneumonia (PCP) hepatitis have been described and are attributable to the use of inhaled pentamidine, which fails to protect extrapulmonic sites from PCP. In addition to PCP, the liver may be the site of infection by the protozoa *Cryptosporidium, Microsporidium,* or *Dicrocoelium dentriticum* or by other multicellular organisms.

Bacillary peliosis hepatis, may be caused by either *Bartonella henselae* or *Bartonella Quintana.*

The significance of a number of the nonspecific findings in the liver are uncertain. In particular, granulomas in the absence of fungal or mycobacterial organisms are common, often prompting concern for tuberculosis. Microvesicular and macrovesicular steatosis is one of the most common nonspecific findings, possibly owing to malnutrition, because

the findings are similar to those seen in patients with kwashiorkor. Massive steatosis has also been observed from antiretroviral drug use (lipodystrophy syndrome).

Biliary tract involvement in AIDS may result in marked liver test abnormalities and right upper quadrant symptoms; jaundice is unusual. A syndrome resembling sclerosing cholangitis with papillary stenosis is well recognized and has been termed *AIDS cholangiopathy*. Patients characteristically develop significant upper abdominal pain in association with marked elevation of alkaline phosphatase, and minimal elevations of bilirubin, AST, and ALT.

Ductular changes may consist of either papillary stenosis alone, sclerosing cholangitis–like lesions alone, a combination of the two, or long extrahepatic strictures. Most series have found papillary stenosis with intrahepatic disease as the most common findings. Ultrasonography or CT detects ductular abnormalities in 77% of those with cholangiographically proven disease, implying that a negative imaging study does not definitively exclude the diagnosis. The etiology in most cases is infectious because *Cryptosporidium,* CMV, or *Microsporidium* may be found in bile, duodenal, or biliary epithelium. For patients with predominantly papillary stenosis, sphincterotomy results in a symptomatic improvement in most patients; alkaline phosphatase may continue to rise, however, probably reflecting progression of associated intrahepatic disease.

Other less common causes of biliary tract disease in AIDS include primary bile duct lymphoma, epithelial angiomatosis, lymphomatous nodal obstruction, Kaposi's sarcoma, and biloma. In addition, chronic pancreatitis or choledocholithiasis may lead to biliary obstruction, although their incidence is not clearly increased in HIV infection.[3,4]

EVALUATION

The initial decision in evaluating the AIDS patient with jaundice, hepatomegaly, or both, is to determine whether the findings are due to intrahepatic or extrahepatic disease. Simultaneous disease in both sites must also be considered. A history of mild jaundice, often in association with fever and constitutional symptoms, is more consistent with intrahepatic disease, whereas symptoms of deep jaundice associated with pain of relatively acute onset suggest extrahepatic disease. Careful review of medications, both prescription and nonprescription, is essential.

Because the clinical history and the finding of symptomatic hepatomegaly are nonspecific, further evaluation is always necessary. Elevations of ALT or AST or both, are common, but neither the pattern nor the extent of elevation of these tests appears to correlate with specific findings in the liver. Significant elevation of the transaminases favors a drug-induced or viral cause. In contrast, marked elevation of alkaline phosphatase correlates statistically with the presence of MAC infection in the liver in AIDS when extrahepatic obstruction is absent. CT scan and ultrasonography should be employed early because they are especially useful in identifying ductal dilation, gallbladder pathology, and focal hepatic lesions.

The indications for liver biopsy for the patient in whom intrahepatic disease is suspected are not well defined. Biopsy is appropriate when symptomatic, treatable disease of the liver is suspected and when a specific diagnosis of hepatic disease is needed.

An extrahepatic cause for jaundice is suggested on CT or ultrasonography by the presence of dilated ducts or other biliary and/or pancreatic abnormalities. Once extrahepatic obstruction is recognized, the possibility of papillary stenosis associated with AIDS cholangiopathy must be considered as well as the possibility of choledocholithiasis or other disorders, depending on the imaging studies. Further evaluation, when indicated, may include endoscopic retrograde cholangiopancreatography (ERCP) if CT or ultrasonography demonstrates extrahepatic biliary ductal dilation. Ampullary and duodenal biopsy specimens or bile and/or biliary cytology (with appropriate staining) collected during ERCP can be examined for the presence of viruses, protozoa, or neoplastic cells. [2,3,4]

Table 1.1 : Major cause of liver injury in HIV infection

Drugs
NRTI, NNRTI, PI Anti microbials Antituberculosis , INH, rifampicin Macrolides : clarithromycin, azithromycin Antifungal : KTZ, itraconazole, fluconazole Anti pneumocystitis carini drugs : TMP-SMX, dapsone
Infections
Viral hepatitis HAV, HBV, HCV, CMV, HSV, EBV Mycobacteria,Mycobacterium avium, M tuberculosis Fungal Cryptococcus, histoplasma, coccidioides, candidia Protozoa Pneumocystitis, toxoplasma,microsporidia, cryptosporidium
Biliary infections
HIV cholangiopathy Acalculouscholecystitis
Neoplasm and vascular lesions
Kaposi sarcoma Lymphoma Peliosis hepatitis
Steatosis and lipodystrophy HCV co-infection Drugs associated

2. REVIEW OF LITRETURE

Since its introduction over 10 years ago, highly active antiretroviral therapy (HAART) has dramatically changed the course of human immunodeficiency virus (HIV) infection by decreasing morbidity and mortality as a result of opportunistic infections. However, HIV-infected patients are now experiencing a wide array of adverse events attributed to the drugs themselves. Liver toxicity is an important example because it carries its own morbidity and mortality. Perhaps more importantly, it often leads to HAART discontinuation

HAART INDUCED LIVER DISEASE

Highly active antiretroviral therapy (HAART) has decreased the morbidity and mortality derived from classical opportunistic infections. As a counterweight to this positive impact, antiretroviral therapy (ART) carries along undesirable effects, which challenge the management of HIV-infected patients to a great extent. Among these, liver toxicity deserves a special attention since it often leads to HAART discontinuation; particularly in hepatitis C (HCV) and/or hepatitis B (HBV) co-infected patients. The mechanisms involved in HAART-derived liver toxicity are not well understood, which makes its management more difficult

One of the toxicities linked to the use of antiretrovirals is the elevation of transaminases. Liver toxicity is a cause of morbidity, mortality, and treatment discontinuation in HIV-infected patients. While several antiretrovirals have been reported to cause fatal acute hepatitis, they most often cause asymptomatic elevations of transaminases

Possible pathogenic mechanisms involved in hepatotoxicity are multiple, including direct drug toxicity, immune reconstitution in the presence of HCV and/or HBV co-infections, hypersensitivity reactions with liver involvement, and mitochondrial toxicity. Other pathogenic pathways may be involved, such as insulin resistance caused by several antiretrovirals, which may contribute to the development of steatohepatitis. The management of liver toxicity is based mainly on its clinical impact, severity and pathogenic mechanism.

CLINICAL IMPACT

With the widespread use of HAART and the availability of more drugs, some of them perhaps more hepatotoxic, HAART-linked hepatotoxicity has been made evident over the past few years. Liver toxicity generates medical visits, work-up exams, and frequent hospital admissions, all of which increase expenses. In addition, hepatotoxicity hampers the maintenance of HIV suppression over time.

In a recent American study, which evaluated the causes of death of HIV-infected individuals, discontinuation of ART due to hepatotoxicity increased from 6% in 1996 to 31.8% in 1998-1999 among those mortalities . More recently, Kramer and colleagues have highlighted the increase in the number of cases of fulminant liver failure in HIV/HCV-coinfected individuals during the HAART era, even after excluding patients with advanced liver disease and adjusting by alcohol intake.

The severity of liver toxicity ranges from the absence of symptoms to liver decompensation; and the outcome, from spontaneous resolution to liver failure and death. Although in a study severe hepatotoxicity with acute hepatic necrosis was present in 2% of HIV+ patients dying due to hepatitis or other liver diseases, mainly among

those with prior liver disease, most cases of liver toxicity is mild-to-moderate and asymptomatic .

Drug liver toxicity has impacted on the recommendations for antiretroviral therapy in certain scenarios. Thus, the use of nevirapine (NVP) has been recommended to be avoided as part of post-exposure prophylaxis regimens. The reason for that was the occurrence of fulminant hepatitis in two cases and severe liver toxicity in 12 other healthy subjects who received a NVP-including HAART regimen after HIV exposure. However, NVP seems to be safe when administered to mother and child as a single dose for prevention of mother-to-child HIV transmission . [3,4]

DEFINITION OF LIVER INJURY

The clinician thinks of liver damage when abnormalities in the liver tests are seen. There is a broad variability among studies in the criteria to categorize the severity of hepatotoxicity. the most accepted one, the AIDS Clinical Trials Group scale of liver toxicity .

According to it, patients with transaminases within normal limits at baseline are considered to develop hepatotoxicity when ALT and/or AST rise above the upper limits of normal (ULN). Severe hepatic injury (the primary study outcome) is defined as grade 3 or 4 change in AST and/or ALT levels during antiretroviral treatment. If AST and ALT grades were discordant, the highest should be used for classification purposes.

DEFINITIONS OF HAART-ASSOCIATED HEPATOTOXICITY: [15]

The AIDS Clinical Trials Group currently uses the following toxicity grading scale:

Patients with normal pretreatment ALT/AST:

Grade 0 hepatotoxicity **<1.25 times the ULN (upper limit of normal)**

Grade 1 hepatotoxicity **1.25 to 2.5 times the ULN**

Grade 2 hepatotoxicity **2.5 to 5 times the ULN**

Grade 3 hepatotoxicity **5.1 to 10 times the ULN**

Grade 4 hepatotoxicity **>10 times the ULN**

There is a separate grading scale for the HAART-associated cholestasis:

Grade 0 cholestasis **<1.1 times the ULN**

Grade 1 cholestasis **1.1 to 1.5 times the ULN**

Grade 2 cholestasis **1.6 to 2.9 times the ULN**

Grade 3 cholestasis **3 to 5 times the ULN**

Grade 4 cholestasis **>5 times the ULN**

For patients with elevated pretreatment ALT/AST, changes are compared to baseline rather than upper limit of normal. Grades 0 and 1 are identical, but grade 2 is associated with ALT/AST 2.6 to 3.5 times baseline, grade 3 3.6 to 5 times baseline, and grade 4 greater than 5 times baseline. [15]

Severe hepatotoxocity is defined as grade 3 or 4 change in transaminase levels, while severe cholestasis, analyzed independent of transaminase levels, is defined as grade 3 or 4 change in total bilirubin.

Many drugs increase γ-glutamyltranspeptidase (GGT) levels. This is often misinterpreted as a marker of liver damage, but the isolated elevation of this enzyme actually reflects enzyme induction. Only when associated with a proportional increase in alkaline phosphatase levels should it be considered as a cholestatic lesion. Bilirubin should not be considered itself as indicator of liver toxicicity it can be elevated due to a variety of reasons, such as hemolysis, fasting and certain drugs (e.g. indinavir and atazanavir). In addition to HAART-derived hepatotoxicity, some other conditions or drugs used in HIV infection, can cause elevations in the levels of liver enzymes and should be ruled out.

INCIDENCE AND RISK FACTORS

The reported incidence of severe liver toxicity after initiating HAART ranges from 2 to 18% . Differences in the study populations, as well as in the methods used probably account for the wide range. In Tables 1 and 2, which summarize the main trials assessing liver toxicity in patients taking antiretroviral therapy, the risk factors are recorded. Hepatitis B and C co-infections [14]

Liver toxicity, especially severe toxicity (grades 3 and 4), is clearly more frequent in HCV and/or HBV co-infected individuals treated with HAART . In one study, a higher risk of hepatotoxicity was found in patients carrying HCV genotype 3 (HCV-3) compared to other genotypes . The clinical implications of this finding are 2-fold. On one hand, the presence of HCV-3 may impact on the selection of HAART regimen, choosing those with less potential for hepatotoxicity. On the other hand, since genotype 3 shows a

higher response to interferon (IFN) and ribavirin (RBV), anti-HCV treatment should be given if no major contraindication is present.

ANTIRETROVIRALS

The results of the studies that have evaluated the risk for liver toxicity associated with the use of particular antiretroviral drugs or families are conflicting. The unbalanced and often insufficient representation of some antiretrovirals in these series, make it difficult to determine with accuracy the role of each particular drug in the development of liver toxicity. In addition, the use of several antiretrovirals combined makes it difficult to ascribe the elevation of transaminases to single drugs. [16]

Protease inhibitors

The phenomenon of hepatotoxicity became more evident after the introduction of ART of high activity, which initially included invariably a protease inhibitor (PI). However, none of the studies has been able to prove the higher potential for liver toxicity of this particular family of drugs. Among the PI, in some studies full-dose ritonavir (RTV) has been found to be more hepatotoxic .In certain cases, RTV has caused fatal acute hepatitis. Several cases of liver toxicity associated with the use of indinavir (IDV) and saquinavir (SQV) have also been reported . Nelfinavir was found to be less hepatotoxic than the other PI anlyzed .

The use of two PI, which often includes RTV at low doses as a booster for the second PI, does not seem to increase the risk of toxicity for the liver. The incidence of liver toxicity with lopinavir (LPV), which is given with low doses of RTV (200mg/day) is low . Atazanavir, marketed more recently, seems also to have a good safety profile regarding the liver, even if used with low-dose RTV . Tipranavir, recently approved, appears to be

more hepatotoxic, most probably because it is given with higher doses of RTV (400mg/day).

Nucleoside analogues reverse transcriptase inhibitors (NRTI)

Some authors have found a lower incidence of hepatotoxicity with lamivudine (3TC) and tenofovir . However, the majority of the NRTI can induce mitochondrial damage, and, therefore, have a potential for the development of liver injury, as it will be explained below . Cases of hepatic failure have been reported in patients taking zidovudine, but didanosine and stavudine have been most often involved in severe hepatotoxicity . Abacavir (ABC) and tenofovir (TDF), with low potential for mitochondrial damage, seem to have a safer profile regarding the liver. In patients with chronic hepatitis B, the removal of 3TC may be accompanied by a flare of HBV replication, translated into an increase in transaminases.

Non-nucleoside analogues reverse transcriptase inhibitors

The risk of liver toxicity associated with the non-nucleoside analogues reverse transcriptase inhibitors (NNRTI) is variable and involves several aspects and mechanisms. Several cases of severe liver toxicity, some of them fatal, in subjects receiving NVP as part of a post-exposure prophylaxis regimen. Likewise, in a trial assessing the NRTI emtricitabine (FTC), a higher incidence of hepatotoxicity was observed among patients taking NVP. Of interest, in both series, the post-exposure prophylaxis and the FTC trial, hepatotoxicity developed early into treatment, and predominated among black women in the FTC study. These data suggest a hypersensitivity reaction causing the liver abnormalities. However, in other reports, the hepatotoxicity of NVP-containing regimens had a later onset (beyond the 4th month),

with an increase in the cumulative incidence over time . Therefore, it looks like there is a second mechanism through which NVP causes liver toxicity, much more common than the hypersensitivity syndrome.

Several retrospective studies have evaluated the development of hepatotoxicity linked to the use of NNRTI. This is especially true in populations with a low prevalence of chronic HCV infection [43]. While some authors have found a higher risk of liver toxicity for NVP compared to efavirenz (EFZ), others have failed to do so . More recently, in a randomized clinical trial comparing NVP twice a day, NVP once a day and EFZ, higher incidences of severe hepatotoxicity were seen in the NVP groups (13.8% once a day and 7.2% twice a day) compared to the EFZ arm . However, only the differences between the once a day NVP arm and the EFZ arm were statistically significant. Taken together, all these data suggest that NNRTI have a greater risk to induce immunoallergic reactions involving the liver soon after initiation of therapy. With prolonged therapy, especially in HBV and/or HCV co-infected subjects, NNRTI have a trend to cause a slight increase in the cumulative incidence of hepatotoxicity, which may spontaneously abate over time. Only in rare occasions is liver toxicity serious. In particular, morbidity and mortality linked to the use of NVP has not been proven to be superior to those of other antiretrovirals.[14,15,16]

Table 2.1 : Liver toxicity of commonly used anti- HIV medications

Dug type	Drug name	Pattern of injury	Comments
Protease inhibitor	Indinavir Saquinavir Nelfinavir Ritonivir	Hepatocellular, distinct histologic pattern including hepatocyte ballooning,Kuppfer cell activation, pericellular zone 3 fibrosis	< 10% of patients have transaminases > 5 ULN Ritonavir inhibits P 450
NRTI	ddC d4T ddl AZT	Microvesicular steatosis	Mitochondrial toxicity manifexting as lactic asidosis
NNRTI	Nevirapine Efavirenz	Hepatocellular	NVP associated with grade 4 toxicity; FDA alert

Other factors

Heavy alcohol intake has also been identified as a risk factor for severe hepatotoxicity in patients taking antiretrovirals . Several authors have identified other risk factors for the development of HAART-derived liver toxicity, such as the prior presence of transaminase elevation, older age , female sex, prior monotherapy , first antiretroviral treatment , lack of response to HAART (only observed at 12 months), and an increase in the CD4 count after HAART initiation . An association between advanced degrees of liver fibrosis and liver toxicity secondary to NNRTI has been recently reported.[17]

Table 2.2 : Common causes and risk factors

Drugs	Malignancy
Obestity	Organ failure (liver)
Coinfection with hepatitis viruses	Cardiovascular disease
Advanced disease	

MECHANISMS OF LIVER TOXICITY

Despite the numerous published studies on antiretrovirals and hepatotoxicity, many unanswered questions still remain, in particular those related to the mechanisms involved. The possible mechanisms involved in the development of hepatotoxicity associated with the use of antiretrovirals are summarized in Fig. 1. It is probable that multiple pathogenic pathways simultaneously concur in some patients, being difficult to identify the exact mechanisms involved in the development of hepatotoxicity.

Table 2.3 : Mechanisms of hepatotoxicity

Direct toxicity
Hypersensitivity reaction
Mitochondrial toxicity
Metabolic abnormalities
Immune reconstitution syndrome in HBV/HCV co-infection

Direct toxicity

Antiretrovirals, as any other drug, can induce direct toxicity in the liver. Drugs metabolized in the liver through the cytocrom pathways may cause liver toxicity when there are polymorphisms in the enzymes . Since many of the antiretrovirals are metabolized in the liver through the cytocrome pathways, idiosyncratic polymorphisms of the enzymatic complexes might lead to significant heterogeneity in drug metabolism, predisposing to the development of hepatotoxicity in certain individuals. Some drugs may potentiate the activation of death receptors and/or intracellular stress pathways.

Hepatocytes promote mechanisms of cytoprotection against the oxidative stress caused by drug metabolism. Heat-shock proteins, induced by various forms of stress including drugs, may exert cytoprotective functions helping to tolerate potentially damaging toxicants . An increase in heat-shock proteins in individuals with polymorphisms may help the liver adapt to and minimize drug toxicity. Anti-oxidation stress mechanisms might explain the spontaneous normalization in the levels of transaminases despite maintenance of HAART, as it occurs with INH. Although still early in its development, pharmacogenomics is a new approach, which may be very valuable to predict the risk for hepatotoxicity in each individual after initiation of ART .

Hypersensitivity reactions

Hypersensitivity reactions are idiosyncratic reactions of the host, not related to the dose of the drug. Sulphas are prototypical drugs inducing these immune-mediated reactions involving the liver. They usually become apparent within the first 4-6 weeks of treatment.

In patients taking NVP, the incidence of symptomatic events involving the liver has been reported to be of 4.9%. In some occasions fatal outcomes were linked to the use of NVP in women with CD4 counts >250cells/mm within 6 weeks after initiation of treatment, due to hypersensitivity reactions. According to a recent analysis, a low body mass index is an independent risk factor for this type of events.

Immune mediated drug reactions seem to involve the generation of neoantigens formed by the reaction of liver proteins with reactive drug metabolites . Assays examining in vitro activation of peripheral blood mononuclear cells against a drug or its metabolites are currently under research. These assays are a promising approach to identify susceptible individuals . On the other hand, individuals with the HLA-DRB1*0101 marker, especially if they have <25% CD4 cells, have been shown to be at greater risk of developing NVP-induced hypersensitivity reactions.

Hypersensitivity reactions have been reported relatively often with NVP and abacavir (ABC), both in HIV-infected patients and in subjects receiving prophylaxis after HIV-exposure , but also with other antiretrovirals such as zalcitabine (ddC) .

Mitochondrial toxicity

It is infrequent but a distinctive type of hepatoxicity that may evolve to acute liver failure. Mitochondrial toxicity is explained in another article. The main feature of the hepatic lesion is the accumulation of microvesicular steatosis in liver cells and mitochondrial depletion. This early lesion may evolve to macrovesicular steatosis with focal necrosis, fibrosis, cholestasis, proliferation of biliary ducts, and Mallory bodies, a clinical picture resembling alcohol-induced liver toxicity, pregnancy steatosis or Reye's syndrome . Of interest, the underlying liver disease does not predispose to this type of

lesion . The ability of NRTI to inhibit mitochondrial DNA synthesis in vitro is in the following order: ddC, ddI, d4T, AZT, 3TC=ABC=TDF . Hydroxyurea, used as coadjuvant treatment with ddI to enhance its activity, seems to increase the toxic effect of some NRTI.

Metabolic abnormalities

Steatohepatitis may cause hypertransaminasemia. Insulin resistance is believed to be the metabolic hallmark of predisposition to non-alcoholic steatohepatitis (NASH). HAART may cause, in the context of the lipodystrophy syndrome, marked abnormalities in the metabolism of both lipids and glucose, including insulin resistance . Mild-to-moderate degrees of steatosis have been found in the liver of patients experiencing HAART-derived hepatotoxicity . Thus, in some patients receiving HAART, insulin resistance and NASH may contribute to the development of liver toxicity. [18]

Studies in HIV-negative individuals find rosiglitazone and pioglitazone can reduce risks associated with metabolic syndrome and can reduce risks for cardiovascular disease associated with insulin resistance and diabetes, as well as in non-diabetics. Preclinical studies suggest that atazanavir may reduce risk for glucose anmormalities in HIV+ patients who have not yet developed glucose abnormalities compared to other PIs associated with risk for glucose abnormalities.

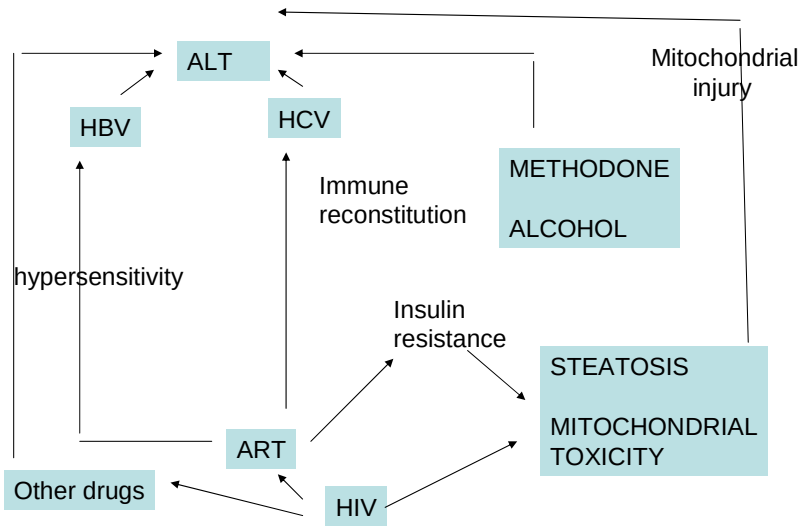
Immune reconstitution in HCV and/or HBV-infected patients

Liver damage induced by chronic HCV and HBV infection is mainly immune-mediated. The immune deficit caused by HIV infection is responsible for the attenuation of the inflammatory reaction in the liver of co-infected subjects. The inhibition of HIV replication with HAART leads to immune reconstitution, and consequently the immune

response to HCV and/or HBV antigens exposed in the liver cell is also restored. Thus, HAART therapy may induce the development of hypertransaminasemia and even symptomatic hepatitis in patients with HCV or HBV co-infection.

The immune reconstitution syndrome as a mechanism of liver toxicity is controversial, but there are several data supporting it. Markers of HCV-specific immune responses (HCV core-specific IGG antibody), T-cell activation and inflammation, have been found to correlate with liver damage and immune reconstitution. Some authors have identified an increase in >50 CD4 cells per mm3 after initiating HAART as an independent risk factor for hepatotoxicity . Liver histology has shown exacerbated viral hepatitis in some patients developing severe hypertransaminasemia while on HAART .

FIG. 2.1 : MECHANISMS OF HEPATOTOXICATY

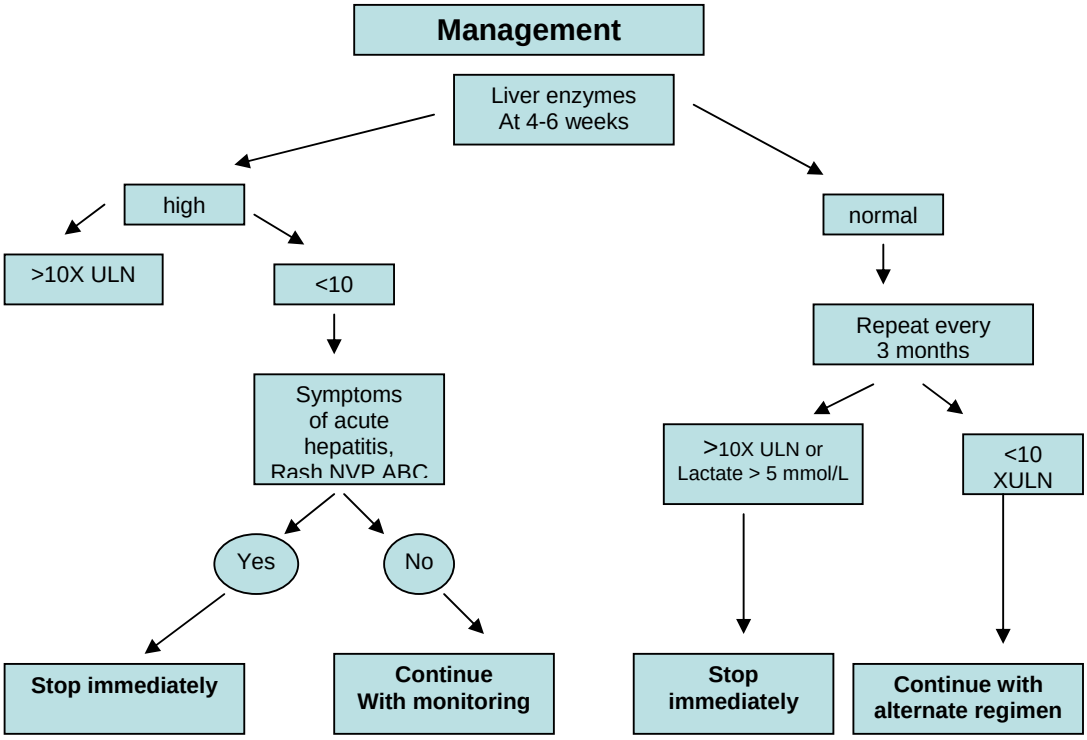


THERAPEUTIC MANAGEMENT

The three main considerations necessary for the management of transaminase elevation after the introduction of HAART are severity, clinical impact and etiologic mechanisms. Suspicion of hypersensitivity reactions or lactic acidosis, and the presence of liver decompensation, are all reasons to stop treatment]. Severe liver toxicity (grades 3-4), even in the absence of symptoms warrants discontinuation of the antiretroviral therapy.

In the remaining cases, decisions should be made on an individual basis. If liver steatosis is present, it is indicated to pursue the removal of predisposing factors. If the patient is taking an antiretroviral with higher hepatotoxic potential, substitution of that particular drug is an available option. Regarding the continuation of the same regimen, spontaneous decrease of transaminases levels has been reported . However, the elevation of transaminases can persist, and the long-term consequences are unknown. In that regard, the finding of an association between NVP use and hepatic fibrosis is worrisome. Nevertheless, this is a single study, and its results should be confirmed by other investigations.

FIG 2.2 : THERAPEUTIC ALGORITHM



Management

Management of HAART-related hepatotoxicity is complicated by the desire of the practitioner to minimize adverse effects while at the same time relying on the chronic use of potentially hepatotoxic agents to prevent life threatening complications from immunodeficiency. Early withdrawal of therapy is recommended for patients who develop symptomatic or severe hyperlactatemia because of a nearly linear association between serum lactate values and mortality. There are anecdotal reports of the use of antioxidants such as riboflavin, L-carnitine, and coenzyme Q-10 (ubiquinone). No controlled studies are available to evaluate these reports. After serum lactate levels and LFTs normalize, reinitiation of retroviral therapy without NRTIs or utilizing NRTIs which have a lower likelihood of causing lactic acidosis, such as lamivudine, abacavir, and tenofovir.[18]

Table 2.4 : Signs and symptoms

Nausea Vomiting Weakness Abdominal pain Diarrhea	Malaise Anorexia Dyspnea Cardiac arrhythmia (fatal)

Table 2.5 : Management

Discontinue ART Deliver adequate oxygen to tissues Reduce O2 demand Mechanical ventilation

Liver biopsy plays an important role in distinguishing between the above mentioned types of HAART-associated hepatotoxicity. By laboratory criteria, most patients with Grade 0, 1, or 2 hepatotoxicity are observed closely, as the benefit of continuing an effective antiretroviral regimen outweighs the risk of severe hepatotoxicity. Patients with Grade 3 or 4 hepatotoxicity or cholestasis are advised to discontinue therapy. Treatment can be re-initiated with a different class of drugs when LFTs have normalized.

3. AIM OF THE STUDY

The study was conducted with the objective of

i. estimate the incidence of drug induced hepatotoxicity in patient receiving HAART therapy for HIV/AIDS

ii. to analyze the risk factors that are associated with drug induced hepatotoxicity in these patients and their clinical profile.

4. MATERIALS & METHODS

About 1523 patients who receive ART were screened and patients with evidence of liver dysfunction were isolated. Sample population was selected as follows.

50 adult patients of both sexes infected with HIV and fulfill the WHO criteria for clinical AIDS receiving HAART for a period of more than 1 month were included in the study.

PATIENT SELECTION

Inclusive criteria

Patients with HIV infection

i. who receive HAART therapy for > 1 month

Exclusion criteria

i. Patients with base line LFT abnormal
ii. No evidence of extrahepatic cause of jaundice
iii. No past history of jaundice
iv. No clinical evidence of liver disease at the institution of HAART
v. Antenatal mothers
vi. Children < 13 years

PROTOCOL

1. All patients who receive HAART therapy who met the above criteria were included in the study

2. The following were noted in each patient

   i. Age

   ii. Sex

   iii. BMI

   iv. Alcohol usage

   v. Smoking

   vi. H/o jaundice in the past

   vii. Drug allergy

   viii. Diabetes

   ix. Tuberculosis (pulmonary/ extra pulmonary)

   x. Mode of acquisition of HIV

   xi. Nadir CD4 count

   xii. Latest CD4 count

   xiii. Socio economic status

   xiv. Dose of each drug

   xv. Duration of therapy of each drug

   xvi. Drug withdrawal

   xvii. Other adverse reaction with the drug

   xviii. Anti tubercular therapy (ATT)

   xix. Anti fungals

   xx. Antibiotics

3. Base line LFTs were measured

4. Those who have abnormal LFT following were noted

   i. USG

   ii. HBsAg, anti HCV antibodies

   iii. Prothrombin time

   iv. Chest X ray

   v. Complete blood count

   vi. Bleeding time

THE STUDY

Study design	Retrospective Analytical Study
Venue	GGH, Chennai
Duration	6 months

Collaborating departments

ART Clinic, Govt. General Hospital, Chennai

ART Clinic, Govt. Hospital for Thoracic Medicine, Tambaram

ART Clinic, Kilpauk Medical College Hospital, Chennai

Liver Clinic, Govt. General Hospital, Chennai

STASTICAL ANALYSIS

Statistical analysis were made by Oneway ANOVA F test. Difference between variables calculated by Chi- Squared test and Student independent t- test using SSPS software.

RISK FACTORS FOR HEPATOTOXICITY IN PATIENTS ON HAART

PROFOMA

S.NO.

NAME AGE SEX UNIT

OCCUPATION

ADDRESS

CONTACT NO.

HOSPITAL NO

SYMPTOMS

PRIMARY SYMPTOMS

GI SYMPTOMS

Fever

Jaundice

Loss of appetite

Loss of weight

Pruritus

GI bleed

PAST HISTORY

Jaundice

Abdominal surgery

Blood transfusions

Hepatotoxic drug intake

Alcohol

Smoking

Tobacco usage of other means

IVDU

Drug abuse

Marital status

Promiscuity M2F / M2M

SIGNS

General

Weight height

Nutrition BMI

Pallor

Oral lesions

Jaundice

Evidence of liver cell failure

Ascites

Cutaneous bleed

Hepatic encephalopathy

DURATION OF HAART

NO OF DRUGS DOSE

Drugs	Started on	Stopped on	Dose

ATT

Drugs	Dosage	Duration	Comments

OTHER ANTIVIRALS ANTIFUNGAL

ANTIBIOTICS

OTHER DRUGS

CAM

PAST H/O JAUNDICE

ALCOHOL Y/N DETAILS SMOKING

IV DRUG ABUSE

NUTRITIONAL STUATUS BMI

PRETREATMENT LFT SERIAL LFT

HBsAg Anti HCV

CMV HSV DM

5. RESULTS

Baseline data

Total screened	:	1253			
Sample size	:	50	hepatotoxicity	:	3.9%
Males	:	35	Females	:	15
Age	:	17 to 49 years	Mean age	:	31
BMI	:	16 - 30	Mean	:	21.2
CD4 count	:	10 - 185	Mean	:	122.5

LFT

Pretreatment LFT			**Post treatment LFT**	
Range	**Mean**		**Range**	**Mean**
0.7-1.1	0.9	T.BIL	0.9-24.5	
		D.BIL	0.8-20.4	
15 – 42	25.6	AST	77 – 1048	280.1
15 – 43	27.4	ALT	88 – 1138	342
		PT	0 – 10	1.4

HBV co-infection : 10 (20%) M : F 8:2

HCV co-infection: 2 (4%)

Alcohol abuse: 16

Baseline date were analyzed by ANOVA F test and found to have no difference between the variables in all the groups. The samples were well matched.(Table 5.1, 5. 2)

Table 5.1

	REGIMEN						
	ALN		SLN		SLE		
	Mean	SD	Mean	SD	Mean	SD	Oneway ANOVA F_test
Age	30.86	7.58	31.95	9.22	26.22	7.01	F=1.55 P=0.22
BMI	21.23	2.88	20.00	3.32	21.89	4.43	F=1.17 P=0.31
CD4nadir	122.45	28.36	99.00	33.81	92.22	61.93	F=2.89 P=0.07
duration	6.64	4.04	5.21	3.54	3.78	1.64	F=2.24 P=0.11
bilirubin	1.26	1.51	.92	.12	.91	.15	F=0.71 P=0.49
AST	25.64	7.05	25.42	7.43	27.11	7.20	F=0.18 P=0.83
ALT	27.45	7.93	28.32	4.03	30.89	5.21	F=0.97 P=0.39
ALP	130.05	26.69	133.58	29.40	130.22	30.82	F=0.09 P=0.91
Albumin	3.07	.21	3.11	.22	3.17	.28	F=0.55 P=0.58

Considering associations between various treatment regimens

REGIMEN 1 : Zidovudine + Lamivudine + Nevirapine (ALN)

REGIMEN 2 : Stavudine + Lamivudine + Nevirapine (SLN)

REGIMEN 3: Stavudine + Lamivudine + Efavirenz (SLE) and

Liver Function Test (TB,AST,ALT,ALP,PT).

The data were analysed by One way ANOVA F test. There was no statistical difference between different regimens in causing hepatitoxiciy ($p > 0.05$).

Table 5.2

	REGIMEN						
	ALN		SLN		SLE		
	Mean	SD	Mean	SD	Mean	SD	Oneway ANOVA F_test
T_BIL	2.93	1.53	5.88	5.73	3.90	4.52	F=2.62 P=0.08
AST1	234.27	153.14	350.00	296.04	286.00	265.46	F=1.21 P=0.30
ALT1	318.68	184.87	416.79	334.13	292.44	247.54	F=0.99 P=0.38
ALP1	196.59	49.90	242.95	160.40	186.22	86.59	F=1.21 P=0.30
PT_N	1.36	1.84	1.89	2.31	1.11	1.27	F=0.62 P=0.55

Considering Age as an independent factor for elevated liver enzymes, 3 age groups were analysed (A < 30 yrs, B 31- 40, C >40). There was no ststistical significance between age and Bilurubin level, AST, ALT, ALP and Prothrombin time (PT). p 0.22

BODY MASS INDEX (BMI)

Analysis of data with BMI as an independent variable showed strong correlation with elevation of Total Bilirubin, AST, ALT, ALP and PT ($p<0.05$)

Table 5.3

	BMI(<18.5)		BMI(>18.5)		Student Independent t-test
	Mean	SD	Mean	SD	
T_BIL	5.93	6.77	3.43	2.05	t=3.95 P=0.05 significant
AST1	377.38	329.75	245.29	169.62	t=3.53 P=0.04 significant
ALT1	489.00	344.01	286.41	185.20	t=7.37 P=0.007 significant
ALP1	243.13	167.68	197.85	69.79	t=1.83 P=0.18 notsignificant
PT_N	1.63	2.58	1.47	1.60	t=0.07 P=0.03 significant

SEX

Comparing the means of liver function tests with Sex did not show statistically significant correlation (p =0.13) (Table A.)

CD4 Count

The association of hepatotoxicity with different CD4 counts catagories was analyzed. Patients belonging to CD4 count < 100 have significant correlation with TB, AST, ALT, and PT. (p 0.04). The risk of hepatotoxicity correlates well with low CD4 count whereas in groups with CD4 count between 100-150 and > 150 did not show any correlation.

Table 5.4

	CD4nadir						Oneway ANOVA F_test
	<100		101-150		>150		
	Mean	SD	Mean	SD	Mean	SD	
T_BIL	6.12	6.02	3.23	2.33	2.49	0.79	F=3.40 P=0.04 significant
AST1	383.89	306.57	242.17	179.88	181.71	70.87	F=3.12 P=0.05 significant
ALT1	420.00	336.85	311.83	194.66	299.71	221.30	F=1.07 P=0.35 Not significant
ALP1	241.00	155.71	199.63	71.35	178.14	62.77	F=1.12 P=0.03 significant
PT_N	1.89	2.33	1.50	1.79	.57	.79	F=1.21 P=0.01 significant

ALCOHOL

Correlation with alcohol and hepatoxicity did not show any statistical significance. (p =0.53).The risk of hepatotoxicity did not have correlation with alcohol consumption.

HEPATITIS B

Coinfection with HBV increases the risk of hepatotoxicity.The data analyzed show positive correlation with HBV positivity with TB, AST, ALT, PT (p0.001). Patients with HBV infection had higher levels of T.B, AST, ALT and Prothrombin compared to negative group.

On combining HBV coinfection and alcohol abuse there was a positive correlarion with TB, AST, ALT, PT. ($p < 0.03$). (Table 5.4)

Table 5.4

	HBV				Student Independent t-test
	No		Yes		
	Mean	SD	Mean	SD	
T_BIL	3.80	4.50	5.92	2.73	t=1.89 P=0.04 significant
AST1	229.83	203.43	518.50	233.25	t=3.90 P=0.001 significant
ALT1	301.38	239.93	550.70	260.58	t=2.89 P=0.006 significant
ALP1	202.30	114.86	252.50	88.23	t=1.29 P=0.22 not significant
PT_N	1.13	1.79	3.10	1.79	t=3.12 P=0.002 significant

.

6. DISCUSSION

There is an increase in the reporting of drug induced liver injury following ART therapy since 1995 following wide spread usage of antiretrovirals.[12]

The incidence of drug hepatotoxicity has been variously reported. It ranged from 5% to 30% in different series (17 cohorts and 2 metanalyses). Less often they cause steatosis, Lactic acidosis and encephalopathy with mortality rates between 0.1 to 7%

Our incidence is likely to be around 3.9%when grade 3 or 4 injury is taken as cut off limit.

We compared our results with that of the literature available on this issue. Much of the data came from large trials like Amsterdam, CHORUS, ICONA and TARGET which involved more than 5100 patients.[3,4,14,15,16,17]

AGE, SEX AND BMI

Age was not considered to be an individual risk factor in most of the published series. Large trials like Saves[17] failed to demonstrate any correlation with age. Our series also found no correlation to the incidence of hepatotoxicity with age.

Female sex was associated with increased incidence of hepatotoxicity in two of the major trials. Martin-Carbonero[23] et al and Wit et al[16] have shown independently that female sex is an independent risk factor and the risk increases with females who are obese and who drink alcohol. Our data found no correlation of hepatotoxicity with gender. Both sexes had equal incidence of liver injury. However the only mortality in our series happened in a female patient.

Obese patients had a higher risk of hepatic steatosis and liver injury (Carr A et al). Other studies have shown that malnutrition and low BMI are also contributory factors in hepatic injury in Asian and African populations (Sampras K et al). In our series there is a strong correlation with liver injury (Bilirubin level, AST, ALT) with low BMI (< 18.5), but did not have impact on normal BMI. Our study population did not include obese BMI patients and thus could not be compared.

DRUGS AND REGIMENS

Previously number of reports of increased liver injury were attributed to certain drugs like Zidovudine, Nevirapine , full dose Ritonavir. Review of 17 clinical trials between 1991 to 2001 in FDA database attributes risk of liver injury for Nevirapine (NVP) and Efavirenz (EFZ). In 2NN study, post exposure prophylaxis of NVP was associated with severe liver injury and it was recommended to exclude NVP from PEP programs.

Our series included 3 standard regimen (AZT + Lamivudine + Nevirapine , Stavudine + Lamivudine + Nevirapine, Stavudine + Lamivudine + Efavirenz). When these were compared with the incidence of liver injury there was no correlation. All regimens had equal incidence of hepatotoxicity following therapy.[20,25]

ALCOHOL

Alcohol usage did not predispose to severe liver injury in studies by Saves et al,[17] Suikowski et al[13] and Rodriguez-Rosado.[12] However Nunez et al[14] found alcohol has correlation particularly following PI based regimens and in obese females (Martin – Carbonero). Our study did not find any correlation of hepatotoxicity with alcohol usage. However we found that there were significant risk associated with presence of combined factors of alcohol and HBV

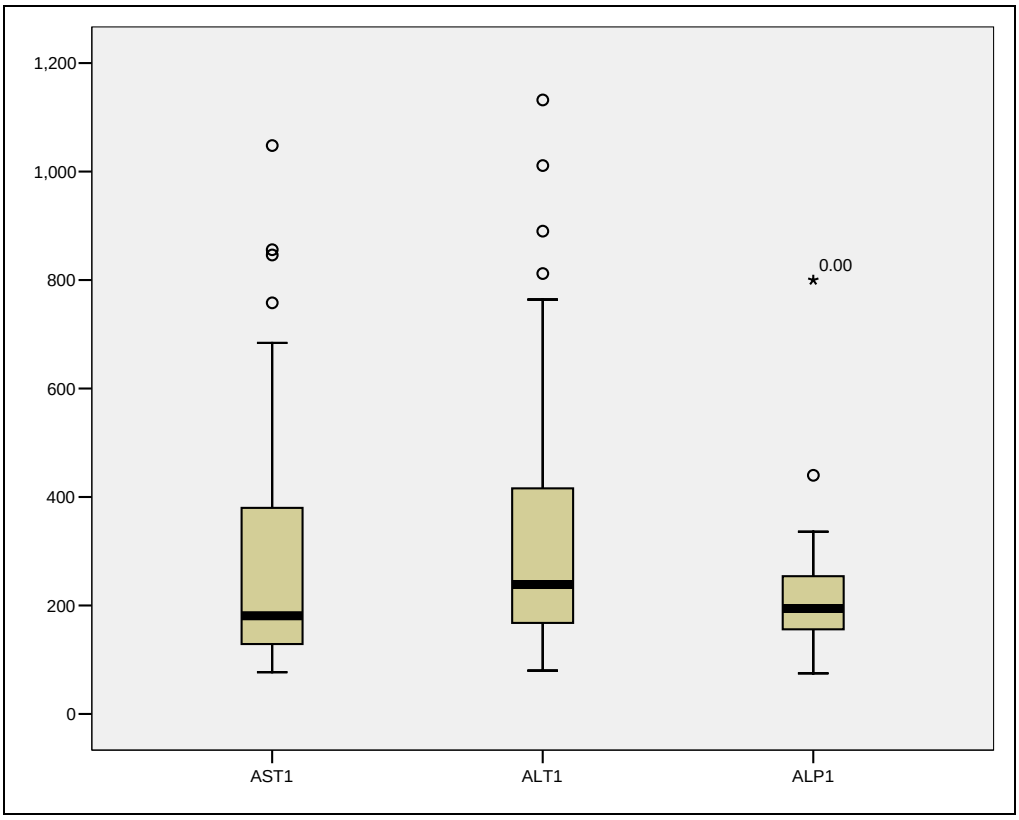


fig 6.1:Alcohol+ HBV Coinfection

HEPATITIS B AND HEPATITIS C

There are atleast 10 studies which show consistent association of liver toxicity with HBV infection. Studies by Saves, Sulkowski[13], Den Brinker, D'Armino, Aceti[26], Wit[16], De Maat have shown that HBV is an individual risk factor.

The risk increased with high viral load, HBeAg positivity and raised baseline AST and ALT. Co infection with HCV is also identified as a contributing factor in these studies. Withdrawal of Lamivudine in HBV positive patients also found to have higher incidence of hepatotoxicity.

In our patients HBV is strongly associated with increased incidence of hepatotoxicity. It has good correlation with Bilirubin, AST, ALT, and Prothrombin time. The incidence of HBV coinfection in the sample is 20% and HCV is 4%.

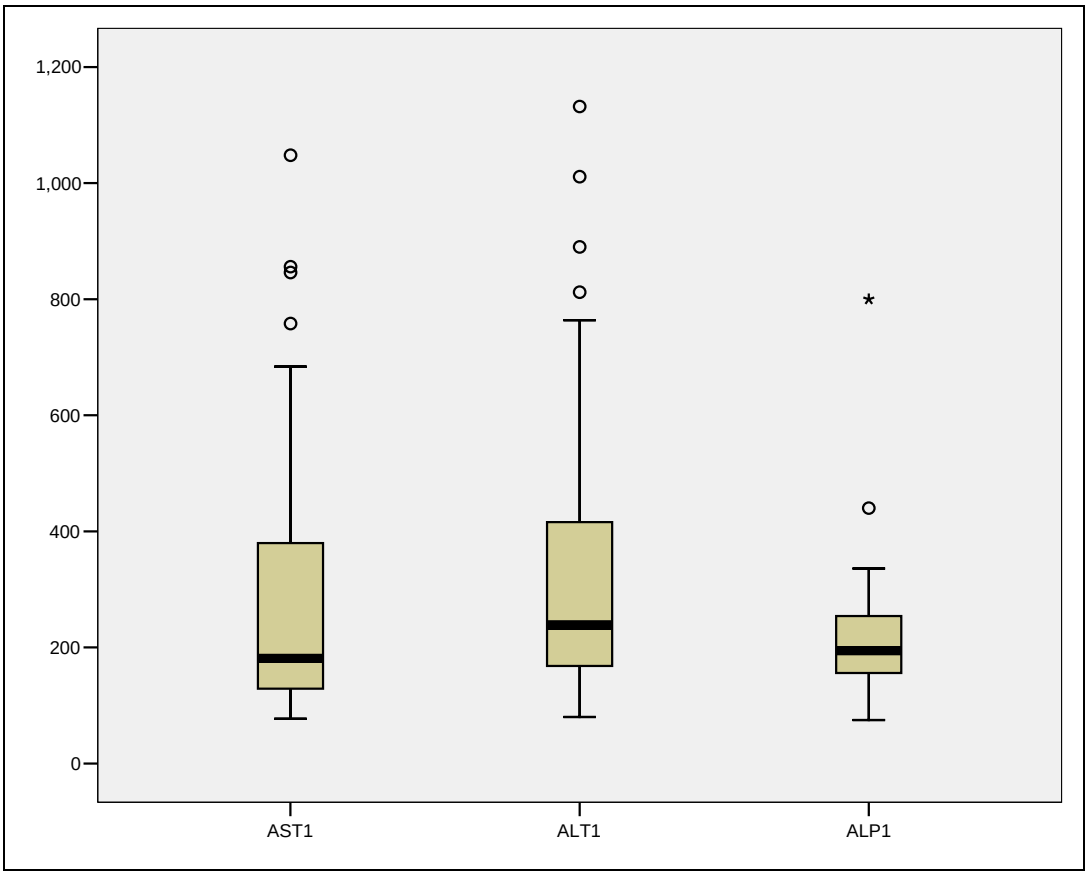


fig 6.2:HBV Co-infection

CD4 COUNT

The correlation with CD4 count and hepatotoxicity was found in our study. Patients with CD4 < 100 cells ran a higher risk of hepatotoxicity and death. This is attributed to profound state of immune suppression and loss of liver regeneration functions. Studies by Saves had similar results.

A study by Sulkowski [13]showed that a rise in CD4 count following HAART predisposes to severe hepatic injury. He attributes this phenomenon to immune reconstitution syndrome where restoration of cell mediated immunity caused aggressive

hypersensitivity reaction to drugs. This phenomenon was also confirmed by other recent studies.[15,16,17]

Most of our patients had CD4 counts moderately elevated after induction of ART and this phenomenon was not observed in our series.

CONSTITUTIONAL SYMPTOMS

We found the presence of rash in about 13.5% of cases preceding liver injury. Fever, malaise, nausea, myalgia and arthralgia occurred in about 33.3%. The incidence was more with Stavudine, Lamivudine and Nevirapine regimen. This observation was not noted in other published series so far.

Presence of peripheral eosinophilia is observed in 13.5% which was lower than observed in Wit et all where he reports it to be around 28%

Presence of rash was neither sensitive nor specific in diagnosing drug induced liver disease following HAART therapy. (fig A 1)

7. CONCLUSIONS

The following were concluded at the end of the study

1. Drug induced liver injury occurs in 3.9% of patients following HAART therapy
2. The risk factors for hepatotoxicity identified were Low BMI, Low CD4 count, HBV co-infection, and combined HBV co-infection & alcohol usage.
3. Age, Sex, alcohol usage alone, various regimens did not have any correlation with the incidence and severity of hepatotoxicity.
4. The occurrence of rash or constitutional symptoms had poor sensitivity and specificity for drug induced hepatotoxicity in these patients.

8. SUMMARY

The study was initiated with the primary aim of finding out the clinical profile and risk factors for the hepatotoxicity in patients receiving HAART therapy for AIDS.

A total of 1523 patients were screened and patients who developed liver injury during ART therapy were selected. About 50 adult patients of both the sexes were included in the study. Records of these patients were analyzed. Further testing was done as needed.

Various parameters were noted.

Statistical analysis was done by Oneway ANOVA F test, Chi-squared test and Student independent t test, in which correlation between various parameters and hepatotoxicity were analyzed.

Results were tabulated and compared with various published series worldwide.

According to our series, the major risk factors for hepatotoxicity were Hepatitis B infection, Low CD4 counts, Low BMI, alcohol abuse in HBV positive patients.

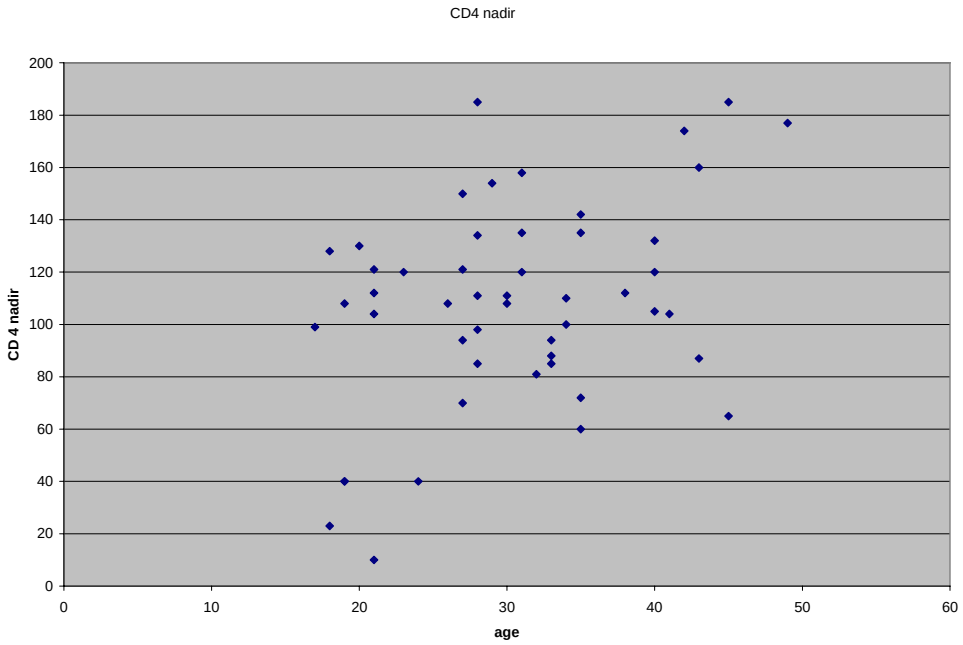
The risk of liver injury was independent of age, sex, various drug combinations.

Clinically, rash and constitutional symptoms occur in less than one quarter of these patients and carry poor sensitivity or specificity.

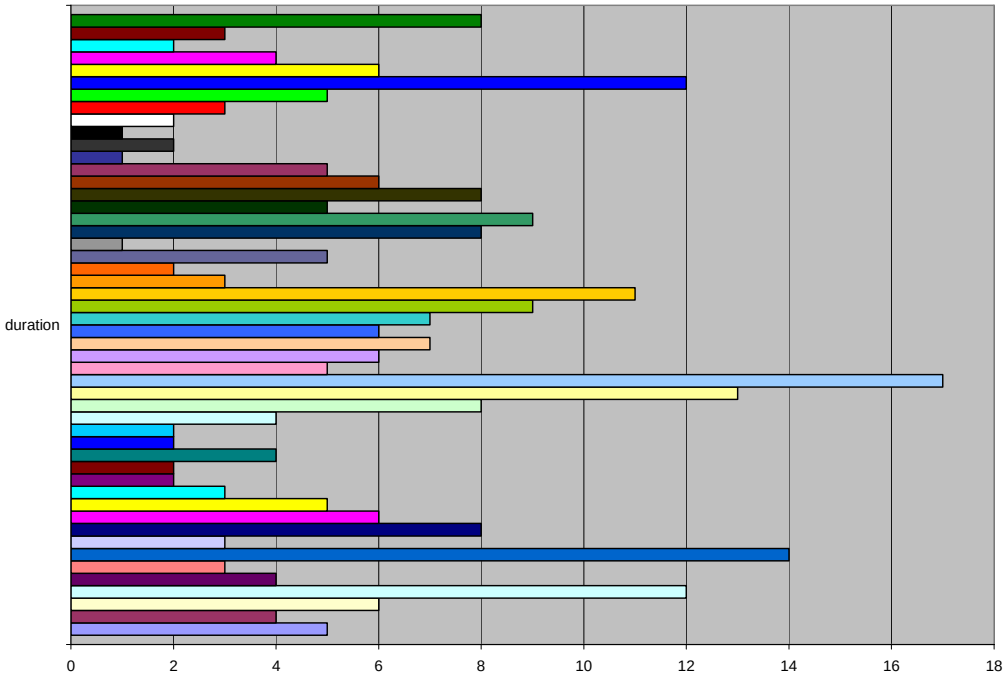
These results were comparable with publishes series from various countries although subtle differences exist

9. APPENDICES

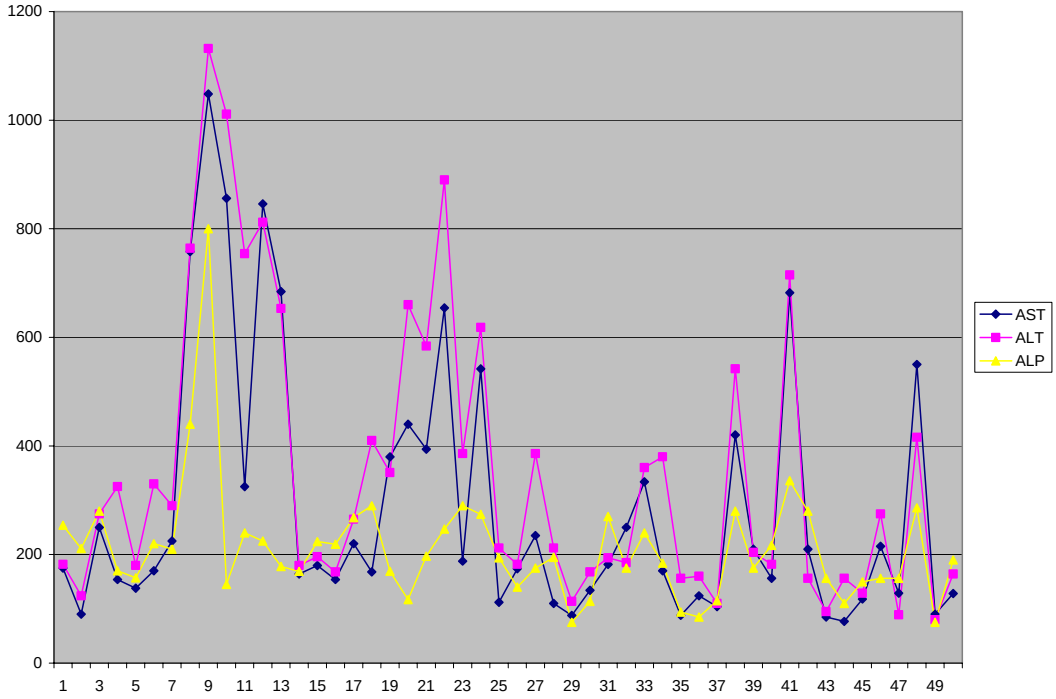
Graph A1: CD 4 counts and Hepatotoxicity



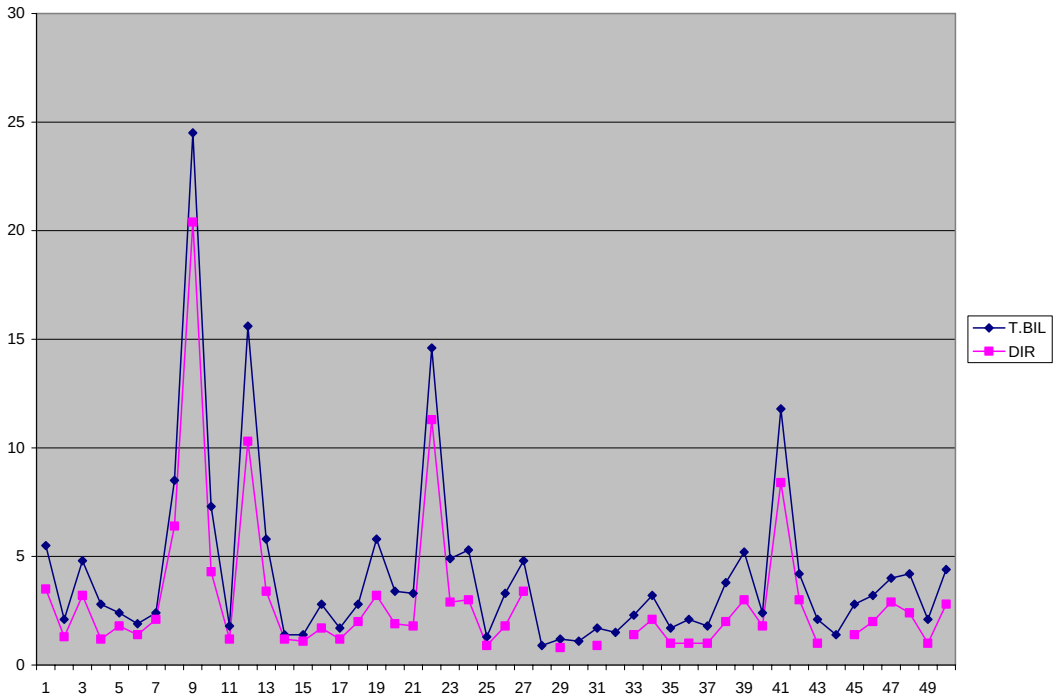
Graph A2 : Duration & Hepatotoxicity



Graph A3 : Liver enzymes values



Graph A4 : Bilirubin Values



Graph A5: Bilirubin & Prothrombin time

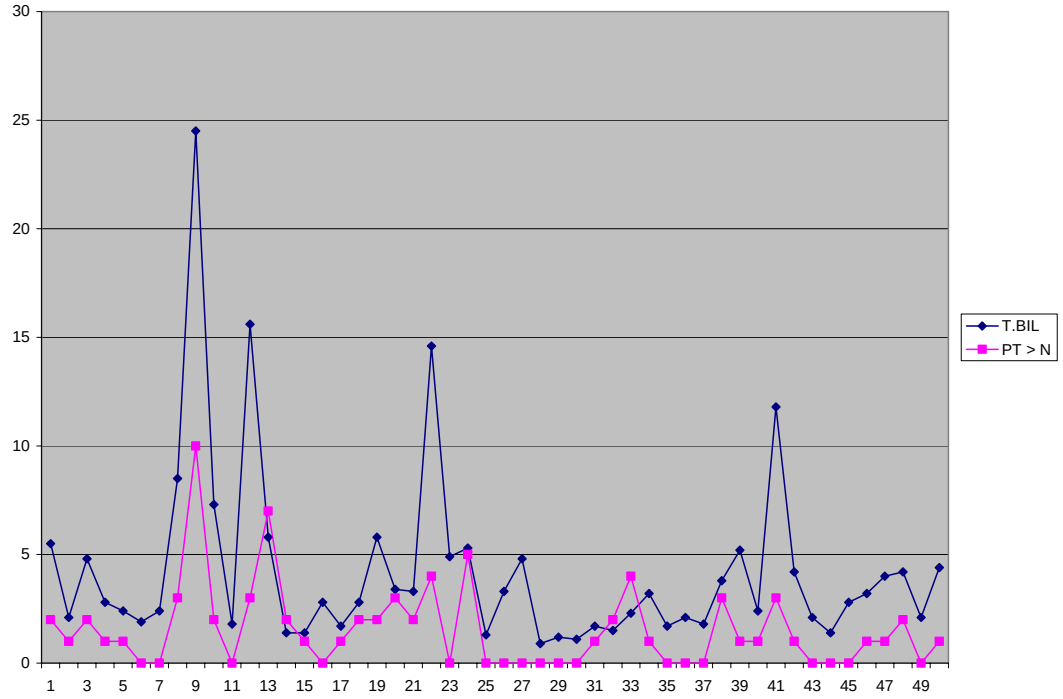


Fig. A1

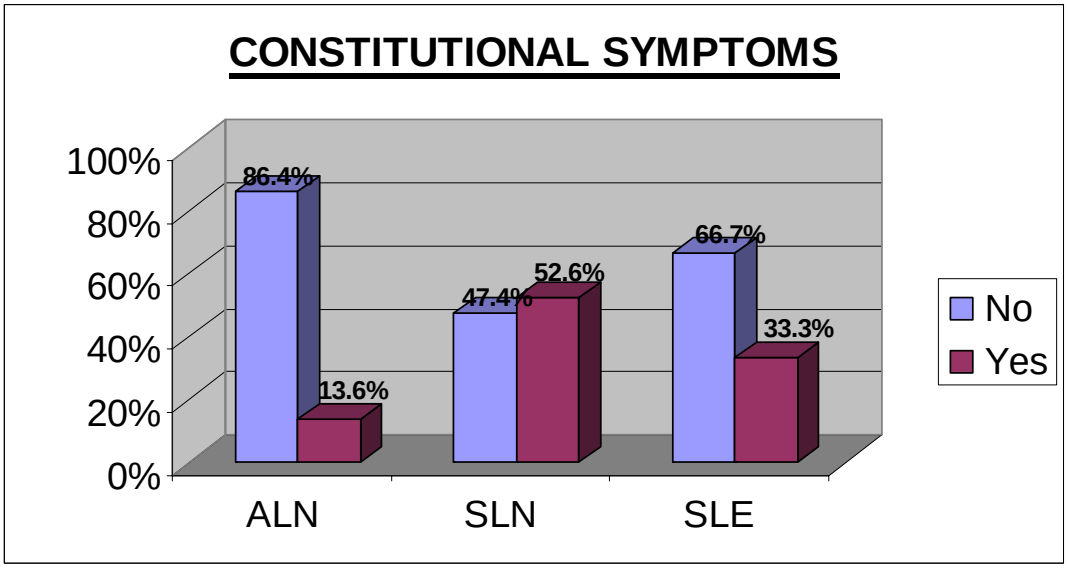


Table A1 : Base line data

	REGIMEN						
	ALN		SLN		SLE		
	Mean	SD	Mean	SD	Mean	SD	Oneway ANOVA F_test
age	30.86	7.58	31.95	9.22	26.22	7.01	F=1.55 P=0.22
BMI	21.23	2.88	20.00	3.32	21.89	4.43	F=1.17 P=0.31
CD4nadir	122.45	28.36	99.00	33.81	92.22	61.93	F=2.89 P=0.07
duration	6.64	4.04	5.21	3.54	3.78	1.64	F=2.24 P=0.11
bilirubin	1.26	1.51	.92	.12	.91	.15	F=0.71 P=0.49
AST	25.64	7.05	25.42	7.43	27.11	7.20	F=0.18 P=0.83
ALT	27.45	7.93	28.32	4.03	30.89	5.21	F=0.97 P=0.39
ALP	130.05	26.69	133.58	29.40	130.22	30.82	F=0.09 P=0.91
Albumin	3.07	.21	3.11	.22	3.17	.28	F=0.55 P=0.58

Table A2 : Baseline Data

		REGIMEN						
		ALN		SLN		SLE		
		n	%	n	%	n	%	Chi-squared test
Sex	Male	16	72.7%	14	73.7%	5	55.6%	$\chi2$=1.09
	Female	6	27.3%	5	26.3%	4	44.4%	P=0.59
alcohol	No	15	68.2%	10	52.6%	8	88.9%	$\chi2$=3.66
	Yes	7	31.8%	9	47.4%	1	11.1%	P=0.16
HBV	No	17	77.3%	15	78.9%	8	88.9%	$\chi2$=0.56
	Yes	5	22.7%	4	21.1%	1	11.1%	P=0.76
HCV	No	21	95.5%	18	94.7%	9	100.0%	$\chi2$=0.47 P=0.79
	Yes	1	4.5%	1	5.3%			

Table A3 : Constitutional Symptoms

		REGIMEN						Chi-squared test
		ALN		SLN		SLE		
		Count	%	Count	%	Count	%	
RASH	No	19	86.4%	16	84.2%	8	88.9%	$\chi2$=0.11
	Yes	3	13.6%	3	15.8%	1	11.1%	P=0.94
EOSINOPHILIA	No	19	86.4%	16	84.2%	8	88.9%	$\chi2$=0.11
	Yes	3	13.6%	3	15.8%	1	11.1%	P=0.94
CONST.S1	No	19	86.4%	9	47.4%	6	66.7%	$\chi2$=7.13
	Yes	3	13.6%	10	52.6%	3	33.3%	P=0.03 significant

Table A4 : Age

	Age						Oneway ANOVA F_test
	<30		31-40		>40		
	Mean	SD	Mean	SD	Mean	SD	
T_BIL	3.74	3.59	4.07	2.86	6.41	8.17	F=1.10 P=0.34
AST1	279.12	214.21	259.29	197.15	387.57	390.70	F=0.75 P=0.49
ALT1	351.23	245.96	291.94	201.24	495.29	405.80	F=1.53 P=0.22
ALP1	189.35	62.09	222.12	89.08	274.00	237.86	F=1.75 P=0.18
PT_N	1.58	1.81	1.12	1.17	2.29	3.50	F=0.92 P=0.41

Table A5 : BMI

	BMI(<18.5)		BMI(>18.5)		Student Independent t-test
	Mean	SD	Mean	SD	
T_BIL	5.93	6.77	3.43	2.05	t=3.95 P=0.05 significant
AST1	377.38	329.75	245.29	169.62	t=3.53 P=0.07 Not significant
ALT1	489.00	344.01	286.41	185.20	t=7.37 P=0.007 significant
ALP1	243.13	167.68	197.85	69.79	t=1.83 P=0.18 notsignificant
PT_N	1.63	2.58	1.47	1.60	t=0.07 P=0.96 Notsignificant

Table A6 : Sex

	Sex				Student Independent t-test
	Male		Female		
	Mean	SD	Mean	SD	
T_BIL	4.82	4.91	2.84	1.56	t=1.52 P=0.13 notsignificant
AST1	320.74	259.00	210.13	160.60	t=1.53 P=0.13 Not significant
ALT1	385.03	290.15	272.40	159.90	t=1.41 P=0.16 notsignificant
ALP1	220.31	123.36	193.73	75.56	t=0.77 P=0.44 notsignificant
PT_N	1.71	2.11	1.07	1.44	T=1.08 P=0.24 Notsignificant

Table A7 : Alcohol

	Alcohol				Student Independent t-test
	No		Yes		
	Mean	SD	Mean	SD	
T_BIL	4.43	5.09	3.82	1.93	t=0.47 P=0.64 notsignificant
AST1	288.91	250.33	284.94	218.64	t=0.05 P=0.96 Not significant
ALT1	334.52	270.51	383.71	248.09	t=0.63 P=0.53 notsignificant
ALP1	214.85	122.31	207.47	88.55	t=0.22 P=0.83 notsignificant
PT_N	1.58	2.24	1.41	1.23	t=0.28 P=0.78 Notsignificant

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